INFLUENCE OF ELECTRICITY ON MICRO-ORGANISMS

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(WITH TWO FIGURES)

The influence of electricity on the higher plants has been studied for many years, and there is considerable literature pertaining to this subject. The writer has carried on investigations in this line for many years, and some of the results have been published from time to time.

Little or no attention, so far as we know, has been given to the study of the influence of electricity on the growth and multiplication of microorganisms, and it is our purpose to present in this paper the results of some of our investigations of the past two or three years.

Microorganisms are favorable types with which to experiment, since they respond very quickly to stimuli, and, as might be expected, the results are more pronounced than is the case with the higher plants, where growth is relatively slower. The investigations given in this paper were made on microorganisms common to water, milk, and soils, and some experiments were made with yeast. In some instances the natural flora common to water, milk, soils, etc., was used, and in others we experimented with pure cultures.

Influence of electricity on bacteria in water

Our first experiments with the influence of electricity on microorganisms were undertaken in connection with those common to water, and were designed with the object of rendering stagnant water more wholesome by a system of electrical treatment. Our studies had not extended very far, however, before we found that, instead of being decreased by means of this treatment, the bacteria increased enormously, especially when weak currents were employed. In this series we made use of the natural bacterial flora of water, while in others isolated species were experimented with. The experiments were made in glass jars, in some cases those of rectangular form being used, and in others a wide-mouthed bottle. For the purpose of measuring currents we made use of a Weston milliammeter and the

usual bacterial methods were employed throughout these tests. All platings were made in Petri dishes in standard agar-agar, and the usual dilution methods were followed. The agar cultures were incubated at the usual temperatures, but the experiments were conducted at room temperatures in most cases, which ranged from 60° to 70° F.[1]

TABLE I

Showing the influence of electrical stimulation (galvanic currents) on the bacteria in water. First cultures made 24 hours after treatment.

Date of making culture	Number of bacteria in 1^{cc}	
	Normal	Treated
June 19........	3463	43,642
" 20........	3435	108,785

The experiment shown in the preceding table was made with two rectangular jars with approximately the following interior dimensions: height 16^{cm}, width 12^{cm}, diameter 4^{cm}. These jars were filled with water obtained from a pond contaminated to a greater or less extent with sewerage. One was a normal or untreated jar, and the other contained electrodes composed of copper and zinc respectively, which were connected with wires and generated a current. The electrodes were of the same diameter as the jar, and one was placed in each end. A jar of this type constitutes a galvanic element (water cell), although the strength of current produced in this case is very weak, averaging about 0.1 milliamperes. Samples of the water were plated in agar-agar 24 hours after treatment. On the second day, however, the experiment was discontinued. The results given in table I show

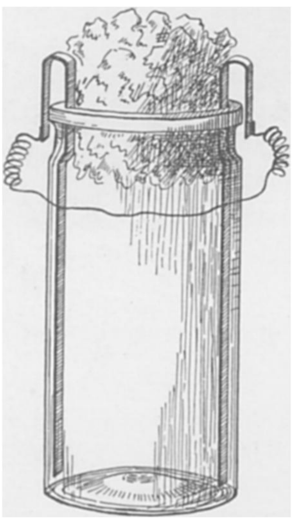

FIG. 1.—Jar, provided with cotton plug and copper and zinc electrodes, used in electrical experiments with milk and water.

[1] In carrying on these experiments the writer is under special obligation to Mr. N. F. MONAHAN, a former assistant in our laboratory, who supervised most of the details of the work.

considerable increase in the number of bacteria in water resulting from electrical stimulation.

The number of organisms in the electrically treated jar increased from about 3000 to 43,000 on the first day, and to 108,000 on the second day.

TABLE II

Showing the influence of electrical stimulation (galvanic currents) on *Pseudomonas radicicola*. First cultures made 24 hours after treatment.

Date of making culture	Number of bacteria in 1^{cc}	
	Normal	Electrical
January 23	6,000	15,000
January 24	50,893	3,178,246
January 27	52,741	4,287,002
January 31	50,217	5,210,112
February 4 ...	50,217	425,000
February 8 ...	42,112	10,200
February 12 ...	41,110	50,000
February 16 ...	35,000	4,000

TABLE III

Showing the influence of electrical stimulation (galvanic currents) on *Bacillus megaterium* DeBary. First cultures made 24 hours after treatment.

Date of making culture	Number of bacteria in 1^{cc}	
	Normal	Electrical
February 25 ...	11,000	243,000
February 28 ...	21,000	3,462,000
March 4......	25,400	5,600,000
March 8......	20,000	4,566,400
March 12......	32,000	7,650,000
March 16......	10,000	243,000
March 20......	35,000	500,000
March 24......	22,000	22,000

The experiments given in the preceding tables were made in wide-mouthed jars approximately 10^{cm} in diameter and 21^{cm} high (*fig. 1*). Those containing the electrically treated water were provided with electrodes made of copper and zinc, which were connected with a wire as in the last experiment. The electrodes were about 4^{cm} wide and long enough to extend over the lip of the bottle. The strength of current developed in this galvanic cell was about 0.3 milliamperes, and it remained very constant throughout the experiments. The

jars in every case were provided with cotton plugs and the whole outfit was sterilized before using. In these experiments pure cultures were used, and the medium, in this case water, was also sterilized before being inoculated. In one series (table II) the jar was inoculated with *Pseudomonas radicicola* (Beyerinck) Moore, from alfalfa; while in the experiments shown in table III the jar was inoculated with *Bacillus megaterium* DeBary, 1^{cc} of a liquid culture medium being used to inoculate the jars in each case. Special care was taken to inoculate the normal and treated jars with the same number of organisms. An examination of the tables II and III will show that there was a marked increase in the number of bacteria during the first few days as a result of electrical stimulation. The maximum in one case was 52,000 for the normal and 5,000,000 for the treated; in another case 32,000 for the normal and 7,000,000 for the treated. It will be noted that the subsequent decrease in the number of the organisms was very marked in the electrically stimulated cultures, a feature due to the accumulation of zinc oxid in the jar, which is always present as a white precipitate in galvanic cells of this type. The presence of zinc oxid in water formed by the action of even comparatively weak currents is toxic to bacteria, and the same toxic effect is well illustrated in galvanotropic experiments with roots. Some of our experiments which were made in much smaller jars failed entirely, as the smaller volume of water employed became concentrated so quickly with this substance that a toxic effect on the organism occurred very shortly after inoculation. Some of the precipitate obtained was dried and dissolved in flasks containing sterilized water. The jars were then inoculated with *Bacillus megaterium* DeBary, with the result that very little increase in the number of bacteria occurred where a 2 per cent. solution of this prepared precipitate of zinc oxid was used, and a 10 per cent. solution apparently killed all bacteria. In both of the experiments enumerated there occurred a slight falling-off in the number of organisms in the normal or untreated cultures. This is of common occurrence, however, in standing water, or even in soils under certain conditions.

The strength of current developed in these experiments (0.1 and 0.3 milliampere) was very constant, and from the results obtained it is evident that it acted as a marked stimulus. A large series of

experiments made by us on the higher plants has shown that this current strength is very close to the optimum, and in all probability the optimum current strength for bacteria would differ little if any from that of the higher plants. We have employed this method of electrically stimulating bacteria and have enormously increased the number of organisms in cultures containing the legume Pseudomonas, which is used in inoculating soils.

Some experiments were also carried on at the same time relative to the influence which electrical stimulation might have upon nitrogen fixation, but the results are incomplete and will not be given in this paper.

The influence of electricity on bacteria in milk

The purpose of our experiments in this series was similar to that in the experiments made with water; that is, to determine the effect of electrical stimulation on the microorganisms in milk. Our object, however, was not only to ascertain the effects of optimum currents, or at least those approximating the optimum on the bacteria of milk, but to observe the effects of strong electrical charges.

Milk constitutes an excellent medium for the multiplication of bacteria and is well suited in some respects to experiments of this nature. The experiments given in tables IV and V were conducted similarly to the ones shown in the preceding series; that is, the bacteria were stimulated by galvanic currents and the same size culture jars were used (*fig. 1*). About 1.5 pints of unsterilized milk were placed in each jar and a milliammeter indicated the strength of current to be approximately 0.3 milliampere in the electrically treated samples. The jars were provided with cotton plugs and were sterilized before being filled with milk. The usual dilution methods were followed and the standard agar-agar was used for plate cultures. In practically all instances the counts are averages of three and four plates. Platings were made of the milk at the beginning of the experiment, that is, before being electrically stimulated; therefore these counts, which are averages, answer for both the treated and untreated cultures.

The results of electrical stimulation on bacteria in milk are shown in the experiments given in tables IV and V, but since milk sours and curdles badly in a few days it was necessary to limit the duration

of the experiments. In the normal or untreated samples the increase in the number of organisms in one experiment was from 143,000 to 6,000,000; while in the treated samples the number reached 94,000,-000. In the experiment shown in table V the normal increased from 118,000 to 4,000,000; while the electrically treated reached 83,000,-000. The results are more striking than those obtained by the treatment of water, as might be expected, since there was more food available for the use of the organisms in the latter series.

TABLE IV

Showing the influence of electricity (galvanic currents) on the bacteria in milk

Date of making culture	Number of bacteria in 1cc	
	Normal	Electrical
May 16, 2 P. M.	143,395	143,395
May 17, 9 A. M.	809,112	3,874,421
May 17, 5 P. M.	1,470,441	86,592,600
May 18, 9 A. M.	6,082,542*	94,851,806*

* Milk sour.

TABLE V

Showing the influence of electricity (galvanic currents) on the bacteria of milk

Date of making culture	Number of bacteria in 1cc	
	Normal	Electrical
May 17, 10 A. M.	118,542	118,542
May 17, 5 P. M.	678,333	1,848,806
May 18, 10 A. M.	1,026,533	41,778,766
May 18, 5 P. M.	4,591,500*	83,363,866*

* Milk sour.

Another series of experiments was undertaken to demonstrate the effects of static electricity on bacteria in milk. For this purpose we employed a static machine of the Töpler-Holtz type, which was designed for X-ray work and is capable of producing a spark six inches or more in length. The method of plating, etc., was the same as has been previously described, and the culture jars were of the same type, except that the electrically stimulated jars were covered with tinfoil in the same way as a Leyden jar. The copper and zinc plates were dispensed with, of course, and the milk was charged direct from the static machine. In this series three jars were used:

one normal, one treated with positive, and one with negative charges. The milk in the electrically treated jars was charged with sparks from a static machine in the same way that a Leyden jar is charged, an electroscope being used to determine the nature of the charge. In the milk experiments which follow it should be pointed out that the charges varied considerably.

TABLE VI

Showing the influence of static electricity (positive and negative charges) upon the bacteria in milk. Electrical jars charged with one spark each June 11.

Date of making culture	Number of bacteria in 1cc		
	Normal	Electrical (positive charge)	Electrical (negative charge)
June 11.............	8,342	8,342	8,342
June 12.............	69,000	196,300	210,000
June 13.............	568,000	1,240,000	1,367,500
June 14.............	1,213,000	16,432,500	19,374,600
June 15.............	9,876,400	70,500,000	79,600,000
June 16.............	27,432,000	153,461,000	131,540,000
June 17.............	190,500,000*	267,000,000*	233,330,000*

* Milk curdled.

TABLE VII

Showing the influence of static electricity (positive and negative charges) upon the bacteria in milk. Electrical jars charged with 10 sparks each June 2.

Date of making culture	Number of bacteria in 1cc		
	Normal	Electrical (positive charge)	Electrical (negative charge)
June 2, 10 A. M	565,000	565,000	565,000
June 2, 5 P. M.......	1,173,000	597,666	624,333
June 3, 10 A. M.......	19,057,000	23,443,666	18,088,333
June 3, 5 P. M........	107,440,000	151,516,000	125,592,000
June 4, 10 A. M.......	201,413,333*	287,380,000*	212,816,666*

* Milk sour.

In the experiment shown in table VI one large spark was given each electrically treated jar, one being given a positive and the other a negative charge, and in the one shown in table VII the number of sparks was increased to 10. The electrical treatment, the results of which are shown in table VI, where the milk was charged with a single spark and cultures made every 24 hours, gave rise to a decided acceleration. This acceleration was perceptible at the time of the

first plating and continued throughout the experiments, which lasted six days.

The results shown in table VII indicate that the treatment given caused less acceleration. In this case platings were made at shorter intervals. The first plating was made seven hours after stimulating; at this time scarcely any acceleration was shown, which indicated the possibility of the death of the organisms by treatment, while during the same period the normal nearly doubled in the number of bacteria. The later platings, however, made at 24, 31, and 48 hours respectively after stimulation, showed a greater increase than that given by the normal, but the ten charges given in the latter experiment were evidently too strong to obtain the optimum results. In both the experiments enumerated (tables VI and VII) the milk was kept on ice.

TABLE VIII

Showing the influence of static electricity (positive and negative charges) upon the bacteria in milk. Electrical jars charged with 100 sparks each.

Date of making culture	Number of bacteria in 1cc		
	Normal	Electrical (positive charge)	Electrical (negative charge)
May 31, 10 A. M.	528,000	528,000	528,000
May 31, 5 P. M.	1,546,000	568,333	601,100
June 1, 10 A. M.	24,885,344	1,323,333	916,666
June 1, 5 P. M.	164,033,000	2,144,600	2,133,460
June 2, 10 A. M.	225,103,632	15,068,333	13,631,090
June 2, 5 P. M.	200,500,000	84,654,000	45,612,000
June 3, 10 A. M.	149,930,000	102,032,420	83,533,220

In another experiment (table VIII) the number of sparks was increased to 100 and the electrically treated jars were charged at 10 A. M. and 5 P. M. each day for a period of three days, with the result that a decided inhibitory effect was noticed after the first treatment, followed by a less increase for the succeeding periods than that given by the normal, and on the third day, when the experiment was discontinued, the electrically treated milk showed a smaller number of bacteria than the normal, thus showing that electrical charges were too strong for the maximum development of the organisms.

A much greater inhibitory effect is shown in table IX, where 100 sparks were applied at more frequent intervals—10 A. M., 1 P. M.,

and 5 P. M. respectively for two days. At the close of the experiment there was little or no difference between the normal and the treated samples. The subsequent charges, however, failed to prevent an increase in the number of bacteria.

TABLE IX

Showing the influence of electricity (positive and negative charges) on the bacteria in milk. Electrical jars charged with 100 sparks each 10 A. M. and 1 P. M. and 5 P. M. each day.

Date of making culture	Number of bacteria in 1cc		
	Normal	Electrical (positive charge)	Electrical (negative charge)
July 3, 10 A. M.	5,580	5,580	5,580
July 3, 1 P. M.	21,440	381	404
July 3, 5 P. M.	78,533	380	487
July 4, 10 A. M.	268,500	5,664	10,367
July 4, 1 P. M.	863,830	10,806	26,990
July 5, 10 A. M.	17,800,000*	19,180,000*	19,910,000*

* Milk sour.

TABLE X

Showing the influence of electricity (positive and negative charges) on the bacteria in milk. Electrical jars charged with 50–100 sparks each every hour (10 A. M. to 5 P. M. inclusive).

Date of making culture	Number of bacteria in 1cc		
	Normal	Positive	Negative
July 6, 10 A. M....	219,250	219,250	219,250
July 6, 3 P. M....	1,000,000	481	266
July 6, 5:30 P. M....	10,655,000*	11,522*	935*

* Sour.

As it was one of the objects of these experiments with milk to determine the maximum stimulus, the charges were increased. For this reason the experiment shown in table X was made, where an even more marked falling-off in the number of organisms is shown. The experiment lasted only a few hours, and was not continued on account of the souring of the milk.

The hourly charges of 50 to 100 large sparks, however, did not entirely prevent the organisms from developing, but the falling-off from over 200,000 to a few hundred in 1^{cc} was significant.

In another experiment, where only one series of heavy charges was given, the number of organisms decreased from 31,000 to 35 one-half hour after treatment; while on the third day the number in the normal was 29,000,000 against 1,000,000 of the treated. Further experiments of a similar nature were made, but in no case were we able to prevent the subsequent appearance of organisms.

The immediate falling-off in the number of bacteria when strong and frequent charges from a static machine were employed would point to the conclusion that the electrical treatment destroyed the bacteria, but what effect it may have had on the spores was not determined. The ultimate increase in the number of organisms in every case after treatment, where heavy charges were used, is also significant, and may be explained on the supposition that strong electrical charges did not affect the spores; in other words, the immediate falling-off in numbers resulting from excessive stimulation may be due to the destruction of the vegetative forms, and those which did appear in the agar cultures may have developed entirely from spores which were possibly not affected detrimentally by treatment. Furthermore, there is a possibility that the strong static charges might induce a tendency in the organisms to spore formation, and the spores, not being affected by the heavy charging, would germinate in the agar cultures.

In the experiment shown in table IX, where a considerable falling-off in the number of organisms took place at first, which was followed by subsequent increase, there was a long period in the night when no stimulation was applied, and spore formation may have taken place; but in the experiment shown in table X the period elapsing between treatments was only one hour, yet the number of bacteria in one instance increased very perceptibly; namely, in the positively charged culture. On the other hand, if the case is one of inhibition or suppression of the vital processes only, we should expect this effect to be lost in the period elapsing between plating and the counting of the colonies. The possibility of accommodation or adaptation of the organisms to intense stimulation is also not out of the question, as this frequently occurs. The roots of certain plants, for example, can grow and develop in water saturated with poisonous gases if given an opportunity to adapt themselves to these extreme conditions,

whereas these plants, if developed normally and placed under these conditions, would suddenly collapse. Our experiments do not furnish sufficient data to determine which of these views is correct. The maximum stimulation would undoubtedly be different for spores than for bacteria existing in the vegetative stage, as it is well known that those in the vegetative stage succumb more readily to heat than those in the spore form. The effects of electricity on spores can best be determined by experiments with pure cultures possessing certain characteristics, rather than those of heterogeneous types such as characterized the flora of the milk with which we experimented.

In our experiments with milk and water, where galvanic currents were employed, the stimulus was constant, whereas when only one shock or a series of shocks was given the organism from a static machine the charges soon disappeared, although the effects of electrical stimuli of brief duration give rise to decided reactions.

It should be pointed out that an increase of even twenty fold in the number of organisms in a given solution at the outset would make a vast difference in the number a few days later, even if the same subsequent rate of increase followed in both the normal and treated cultures. The amount of available food supply in a solution, however, is limited, and in the end there is often little difference in the number of organisms present in any treated or untreated series.

Undoubtedly the use of strong electrical currents is capable of destroying bacteria and preventing milk from deteriorating, although other methods of electrical treatment would probably prove more satisfactory than those which we employed. In some tests made of electrically treated milk we found that souring was delayed. It is well to note in passing that there appears to be a difference in the effects of the positive and negative charges; for example, if a comparison is made of the averages of the last counts in tables VI to X inclusive, it will be found that the milk treated with positive charges gave a larger count than that treated with negative charges. The number of bacteria shown in the positively charged jars was 135,000,000, while that of the negatively charged was 109,918,164. This is what might be expected, since the writer has previously demonstrated in a large series of experiments on the growth of seedlings that positive charges stimulated both roots and stems more than the negative charges.

Feeble electrical currents and small static charges act as stimuli to bacteria in milk, increasing their numbers very perceptibly, and under certain conditions there may be after all some foundation for the old belief that milk sours more quickly during thunderstorms than at other times. Notwithstanding the fact that there may be other conditions during thunderstorms, such as warm and sultry weather, which may accelerate bacterial action, it is not difficult to imagine conditions under which milk might be stored which would subject it to electrical stimulation, thus increasing the number of bacteria and incidentally hastening souring.

The influence of electricity on bacteria in soils

Only a limited number of experiments was made by us relative to the influence of electricity on bacteria in soils. Careful bacterial analyses of soils are tedious operations and the methods followed in these analyses were similar to those recommended and used by CHESTER.[2] In all cases the counts were made on one gram of air-dried soil. In these experiments we made use of wooden boxes $8\times8\times8$ inches, inside measurements, which were filled with fairly good loam free from coarse material. The soil used was more or less compact, very fine sand ($0.01^{mm}-0.005$) predominating. It might be expected that the texture of the soil, as determined by the size of the particles, the amount of organic matter, and plant food, would exert an important influence on the bacterial flora, and the rather fine texture of the soil which we selected for this experiment would give results different from those that would be obtained from a looser soil containing larger particles and having more air space, or one containing a larger amount of organic matter.

In the box electrically treated were placed copper and zinc electrodes, each being 8×8 inches in size, and to these were soldered copper wires which were connected, thus forming with the soil a galvanic cell which furnishes a small current approximating the optimum. The percentage of water in the soil in each box was accurately determined at the beginning of the experiment, and this same percentage was maintained throughout by adding sterilized distilled water. The experiments were carried on in the laboratory under conditions as

[2] CHESTER, F. D., Delaware Agric. Exper. Sta. Bull. 65 : 61–65. 1904.

nearly identical as it was possible to make them, the soil being exposed to the air continually. The cultures were plated in agar-agar and the usual dilution methods were followed.

TABLE XI

Showing the results of electrical stimulation (galvanic currents) on the bacteria in soil.

	Date of sampling	Number of bacteria in 1gm	
		Normal	Electrical
Experiment I.......	July 13	33,470,000	37,930,000
	July 29	28,777,000	32,863,000
	August 11	19,294,000	35,000,000
Experiment II.......	September 14	38,047,000	37,670,000
	October 16	18,720,000	26,384,000

Two experiments are given in this table, the first date of sampling corresponding with the beginning of the experiment. No attempt was made to disturb or cultivate the soil in the boxes, and the surface became more or less compacted by constant watering, which no doubt accounts for the general falling-off in the number of bacteria in all cases. It will be noted that no increase is shown in the number of bacteria in either the treated or untreated soils, although the extent of falling-off in both experiments is less in the electrically treated boxes. Similar experiments were made with soil in the same boxes with static electricity. The boxes in this case were arranged as follows: one box was left normal as before, and the other had 12 wires extending into the soil leading to a metal bulb which was given 100 sparks from a Töpler-Holtz machine once a week. In this case the first samples were taken a few days after electrical treatment; otherwise similar methods were employed as in the preceding series.

In this case there did not occur the same falling-off or decrease in the number of the organisms in either the treated or untreated cultures at the time of the subsequent platings, as in the preceding series. On the other hand, the treated soil in this experiment showed a continual increase in the number of bacteria present at the time of the different platings. This was apparently due to the frequent stirring or cultivation of the soil. The number of organisms, however, showed only a slight increase at the time of the first platings, whereas the

electrically treated increased from 4,000,000 to 27,000,000. The soil in this case was freshly prepared and carefully mixed and contained more organic matter and plant food than the former; neither was there the same tendency for the soil to become badly compacted as in the preceding series.

TABLE XII

Showing the results of electrical stimulation on the bacteria in soil (static electricity).

Date of sampling	Number of bacteria in 1^{gm}	
	Normal	Electrical
July 21.....	1,097,290	4,506,700
July 31.....	960,000	15,208,000
August 7.....	1,960,780	27,756,000

The electrical experiments with soil were not continued, since the details associated with soil bacteriological analyses are laborious. The results of electrical stimulation of soil organisms are not so pronounced as in the case of the water and milk experiments, but the effects are clearly shown in table XII.

Our numerous experiments in growing plants in electrically stimulated soils have demonstrated that considerable acceleration in germination and growth follow when currents of optimum intensity (0.1–0.6 milliampere) are employed, and all the forms of plant life are undoubtedly stimulated in a similar way.

Influence of electrical stimulation on yeast

Some experiments were made in our laboratory for the purpose of observing the effects of electrical stimulation on yeast, in which we endeavored to determine the relative activity of the normal and electrically treated organisms by the amount of CO_2 given off.

As in the preceding series, we used galvanic currents obtained by the use of copper and zinc electrodes and also static charges from a Töpler-Holtz machine. Small bottles were used in the experiments, ranging from 2 to 4^{cm} in diameter and 12 to 16^{cm} high. The electrodes were 2^{cm} in diameter and were placed in the bottle containing the yeast and culture media. In the experiment with static electricity we made use of simple Leyden jars corresponding to those described

in the preceding series. In all instances corresponding jars were employed in the normal and electrically treated yeast, and the conditions were identical so far as they could be made in every way in all cases. From 0.5 to 5^{gm} or more of yeast were placed in each jar in the various experiments, and either a solution of molasses (about 5 per cent.) or a standard nutrient solution was used. The nutrient solution was made up as follows:

	Grams
Calcium nitrate	6.0
Potassium nitrate	1.5
Magnesium sulfate	1.5
Neutral potassium phosphate	1.5
Sodium chlorid	1.5
Cane sugar	125.0
Distilled water	2500.0

This solution was not chosen on account of its being the best adapted for this purpose, but it proved to be satisfactory.

The yeast used in these experiments was from the ordinary commercial yeast cakes, which were cut into cubes and carefully weighed. The yeast was then put into mortars containing the nutrient solution and the cells carefully separated by repeated stirring. After the yeast cells were well separated they were placed in their respective bottles. The mortar was thoroughly rinsed and care was taken to have the same amount of yeast in each. The bottles containing the yeast were completely filled and connected by means of glass tubes to graduated cylinders or burettes containing water, and as CO_2 was given off, the displacing of water was noted at intervals and recorded. The following table gives the results of some experiments with yeast.

The experiments (see table XIII) had a duration of 1.5 hours to 4 days, and in all instances the amount of CO_2 given off was greater in the electrically treated than in the normal or those not stimulated. The experiments were conducted on different days and at different temperatures, most of them being at room temperatures, and as there was no heat on in the laboratory at the season of the year in which many of these experiments were conducted, the temperature was naturally somewhat variable. In *a*, *b*, *c*, *g*, and *h*, however, the bottles containing the yeast were in water baths and were kept under conditions nearer to the optimum for yeast (32–38° C.), hence the per-

centage of CO_2 given off in these was greater for a given period than in some of the others. All of the other experiments were run at room temperatures, which in some instances were fairly good, and in others the room was too cool to expect much activity on the part of the yeast. The small amount of CO_2 given off in some instances is therefore due to the temperature conditions under which the experiment was made, and moreover, since these experiments in some instances lasted four days, there was more or less absorption of CO_2 by water in the graduated cylinders.

TABLE XIII

Showing the influence of electricity on yeast

Experiment	Nature of electrical stimulus	Duration of experiment	Number of cc of CO_2 given off in	
			Normal	Electrical
a	galvanic	2 hours	174	212
b	galvanic	3 days	752	838
c	galvanic	2 hours	189	232
d	galvanic	3 days	540	700
e	galvanic	4 days	20	296
f	galvanic	4 days	50	1200
g	static*	1.5 hours	112	129
h	static	2 hours	74	102
i	static	7 hours	82	120
j	static	3 days	922	1035
k	static	2 days	300	575
l	static	4 days	15	140

* *g* was given 8 sparks; *j* and *l*, 5 each; *k*, 1 spark; *h*, 2; and *i*, 3.

The absorption of CO_2 by water was noticeable when the yeast cultures were left over night, especially when the room temperature was low and the yeast not active. The absorption of water, however, was apparently the same in both the treated and untreated series and did not affect the relative results. In some cases, therefore, a larger amount of CO_2 was given off than is shown by the records in the tables.

Frequent observations and readings were made of the amount of CO_2 given off, but to make the tables brief we have given only the final readings.

The effects of electrical stimulation seem to be more pronounced in the lower temperature experiments than in those where there was a high temperature. In some instances the untreated cultures gave

off very little gas, while the treated, under the same temperature conditions, gave off considerable. The electrical stimulation, under these conditions, appeared to act very much as yeast would if subject to an increase in temperature.

In those experiments where the carbon dioxid gas readings were made at short and regular intervals, we were able to observe the effects of the stimulation on the organisms at different periods.

In some experiments the observations were made every five minutes from the start, and after a number of observations were made and recorded, the stimulus was applied. The results obtained in one of the experiments, in which five-minute observations were made, are given in the curve shown in *fig. 2*.[3] This curve is based on the increased amount of carbon dioxid given off by the normal or untreated; in other words, the amount of CO_2 generated by the normal in this case would be represented by the base line, or it would be equivalent to zero. The treated yeast was given two very small sparks from a static machine at 1:50 P. M., or one-half hour after the experiment was started. A temperature of 30–35° C was maintained during the experiment.

The relative amount of CO_2 given off by the electrically treated and untreated yeast before the stimulation was applied showed quite a uniform activity on the part of the yeast. Following the latent period, which is usually of 15 to 25 minutes' duration, the results of the treatment were manifested in considerable acceleration in gas production. This reached its maximum effect at 3:20 P. M., or 1.5 hours after the stimulation had been applied. Another stimulus consisting of two minute sparks from a Töpler-Holtz machine was applied at 3:45 P. M. and produced a brief reaction, as shown by the curve. The experiment, however, was discontinued at 4:20 P. M.

In the experiments with yeast there was considerable variation in the number and size of the sparks applied to the treated jars, and there appears to be an indication, from the results obtained, that in some instances the electrical charge was too severe. This was noticed in the short-interval readings immediately following the stimulation, and in such cases the maximum acceleration period was more remote

3 We are indebted to Mr. G. H. CHAPMAN, assistant in the laboratory, for supervising this experiment, as well as one other in this series.

from the stimulation period than in this case, where very weak charges were used. A charge of one or two minute sparks from a Leyden jar seemed to cause the most active response on the part of the yeast.

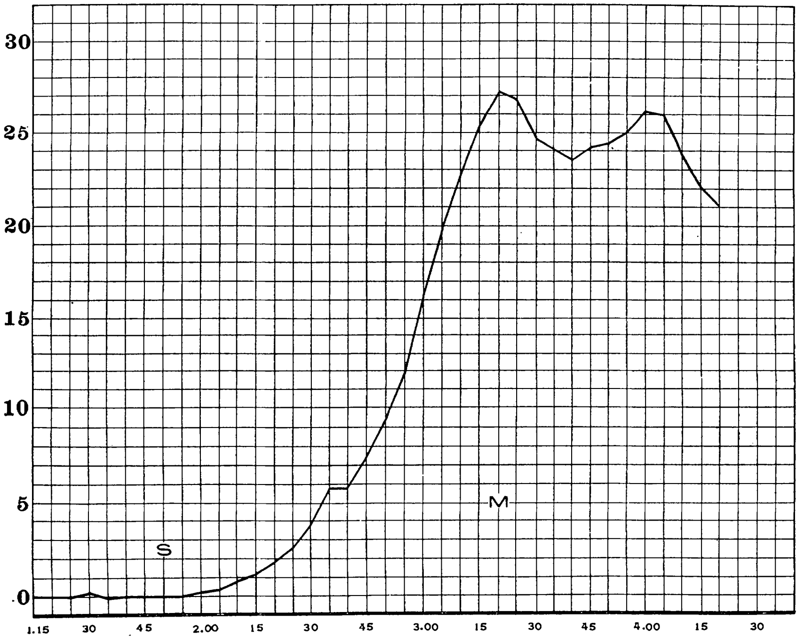

FIG. 2.—Curve representing the amount of CO_2 given off by electrically stimulated yeast; the horizontal lines represent the amount of carbon dioxid in cubic centimeters; the vertical lines five-minute periods of observation; *S*, time stimulation was applied (1:50 P. M.); *M*, maximum effect of stimulus.

Discussion of results

In considering the results presented here it should be pointed out that little attempt was made to ascertain the strength of the current necessary to produce the best results, and the static charges employed differed in number and intensity. We have already demonstrated in a large series of experiments[4] with the higher plants that the optimum current for germination of seeds and growth of seedlings is not far from 0.1 milliampere, and in our study of the effect of static

[4] STONE, G. E., The influence of current electricity on plant growth. Ann. Rep. Hatch Exper. Sta. Mass. Agric. Coll. **16**: 13–31.

charges on the germination of seeds and growth of seedlings we have observed that a very few minute sparks from a static machine caused the most marked stimulation.

In regard to the influence of atmospheric electrical potential on growth, MONAHAN[5] found that when the air in a glass case is charged to a potential of about fifty volts, better results were obtained than when a higher potential was used. We endeavored, therefore, in these experiments to make use of current strengths approximating the optimum, or that strength which gave the best results in our previous investigations with various organisms, except in those cases where strong static charges were given to milk for the purpose of ascertaining the degree of stimulation which would kill the organisms.

The results obtained from these researches suggest many lines of work which might be followed, but we are obliged to discontinue them for the present. Electricity undoubtedly, in one way or another, plays a very important rôle in plant life. Seed germination and growth of seedlings are greatly accelerated by feeble currents, but, unlike amides and enzymes, they are incapable of affecting the germinating capacity or of regenerating, as it were, the life in the seeds. The roots of the higher plants exist in a medium which is charged negatively, and the electrical potential of the air is often quite high within the limits of large trees. The electrical potential under the foliage of a tree is less than that at corresponding heights in the free atmosphere.[6] When, however, there is no foliage, the electrical potential under the branches of trees corresponds to that of the free air at equal heights, and there is reason to believe that the apices of leaves are merely so many points for the gathering and discharge of electricity. Minute currents of electricity exist in plants, and it is known that during certain periods trees discharge sparks from the apices of the leaves, and trees may tend to equalize differences in potential existing between the earth and air. Rain drops in falling become electrically charged, and as they gather microorganisms in their descent through the air, these also probably become affected. The remarkable influence of rain upon

[5] MONAHAN, N. F., The influence of atmospheric electrical potential on plants. Ann. Rep. Hatch Exper. Sta. Mass. Agric. Coll. **16**: 31–37.

[6] STONE, G. E., AND MONAHAN, N. F., Ann. Rep. Hatch Exper. Sta. Mass. Agric. Coll. **17**: 13–31.

vegetation cannot be satisfactorily explained, in our opinion, by chemical analysis or by the various other conditions which prevail, and the idea that electrical stimulation plays an important rôle here is not an improbable one. It is also not unlikely that during thunderstorms the bacteria in milk are affected, although two series of experiments made by us in exposing milk in sterilized metal vessels at different elevations, where the electrical potential showed considerable variation, were by no means conclusive.

The effects of electrical stimulation on plant growth resemble more nearly those produced by heat, that is, in the tendency of the plant to assume a rather spindling growth; but this similarity in the method of reacting does not necessarily prove that electricity and heat are identical, since spindling growths in plants occur from other causes. The effects of electrical stimulation do not resemble those induced by light, since light inhibits growth; on the other hand, they more closely resemble the effects induced by lack of light (partial etiolation) and other forms of stimulation which may be produced by various agencies. Electricity stimulates seeds very perceptibly, causing an acceleration in growth, and probably has the same effect on spores, and in this way the number of bacteria in solutions might be increased. The process of cell division of bacteria and the budding of yeast are undoubtedly stimulated by electricity, which would result in an increase in the number of organisms and an acceleration of the metabolic process.

The effects of electrical stimulation, like other types of stimuli, are manifested shortly after application. With a current of optimum intensity a latent period occurs when no effect is discernible, and this is followed by an acceleration in growth and development. The nature of the response is dependent upon the intensity of the stimulus as well as upon its duration; therefore to determine the period of duration of any particular response or its maximum period, the intensity and duration of the stimulus must be taken into consideration. Since the intensity and duration of the stimulus employed in these experiments differed materially, the response periods would also vary accordingly. As regards the manner in which electricity stimulates organisms, little can be said at the present time, and the problem is as difficult of solution as the manner in which light, etc., affect the organisms.

Many theories, however, in regard to the cause of the stimulating effect of electricity on plant growth have been advanced, some of which are hardly worthy of consideration, since they fail to meet the requirements of experiments, and we will not enter into a discussion of them here.

Electricity, like other forms of stimulation, such as light, heat, etc., undoubtedly affects the protoplasm of the plant, which causes certain metabolic processes to become active and accelerated growth results. In plants showing circulation and rotation of protoplasm, e. g., Chara, Nitella, etc., feeble electrical currents induce a more rapid streaming of the protoplasm, which is undoubtedly associated with greater metabolic activity, and it is not at all unlikely that changes of a similar nature take place in other organisms when subject to feeble electrical currents.

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